REPRODUCTIVE SYSTEMS AND CHROMOSOME RACES OF *OXALIS PES-CAPRAE* L. AND THEIR BEARING ON THE GENESIS OF A NOXIOUS WEED[1]

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ABSTRACT

In its native South Africa, *Oxalis pes-caprae* is represented by diploid and tetraploid races; the short-styled, sterile pentaploid race reported as a noxious weed elsewhere is apparently uncommon there. South African plants have trimorphic flowers, but the three morphs usually are not present in equal proportions in natural populations. The diploid and tetraploid races of the species have a well-developed incompatibility system associated with their floral trimorphism. Outside South Africa, the species is represented not only be a fairly sterile short-styled pentaploid, but sexual tetraploids are known as well. Although the latter may have resulted from independent introductions, low levels of sexuality in the pentaploids also could account for the origin of tetraploidy in situ as a consequence of the union of diploid gametes, and may account for the occasional spontaneous appearance of mid-styled plants in populations of the short-styled pentaploid. Pentaploidy likely resulted from the union of an unreduced gamete of a tetraploid with a haploid gamete of a diploid and presumably developed in South Africa. Since weediness is characteristic of many populations of the native diploids and tetraploids in South Africa, pentaploidy per se cannot be implicated in the origin of weediness, nor is there evidence of any aggressive superiority of the short-styled morph in the sexual races in South Africa. Why the pentaploid is such a successful weed outside South Africa, but apparently less successful as a weed than the diploids and tetraploids within South Africa, is unknown.

Oxalis pes-caprae L. is a troublesome and widespread agricultural and garden weed, particularly in areas of the world with a mediterranean climate such as central Chile, the Mediterranean basin, parts of Australia, and California (Salter, 1944; Young, 1958; Munz, 1959; Michael, 1964). The species is native to southern Africa, where it is variable (Salter, 1944), and where it is distributed from Namibia (South West Africa) southward to the Cape region and around the Indian Ocean coast at least as far north as the Knysna area, sometimes ranging well inland. There it occurs as a "well-behaved" native of relatively undisturbed sites as well as a weed, and it is particularly common in vineyards and along roadsides. In southern Africa, the species is tristylous (Fig. 1), but throughout most of its exotic range it is represented by a short-styled form, which is pentaploid ($2n = 35$), and which reproduces asexually via bulbils. In parts of its exotic range, tetraploid and presumably sexual populations also occur, though less commonly than the sterile pentaploid.

In view of the importance of this species as a weed, certain features of the reproductive biology and chromosome cytology of plants originating in South Africa were studied in the hopes of elucidating the events that led to the origin of the aggressive weedy races from the native races.

MATERIALS AND METHODS

Bulbs collected from natural populations of *Oxalis pes-caprae* in the Cape Province region in South Africa in 1970 and 1971 were later grown at Berkeley for chromosome studies. One collection was provided by Sherwin Carlquist. Chromosome numbers were determined by examining microsporogenesis in flower buds of these cultivated plants preserved in 3 : 1 ethanol : acetic acid and stained in acetocarmine. Bulbs collected by Peter Goldblatt from two localities in 1984 provided plants used in an artificial crossing program to determine the presence and nature of an incompatibility system in this tristylous species. These localities are an abandoned farm near Noordhoek, Cape Peninsula, and a vineyard at Rustenberg, near Stellenbosch, Cape Province. The two localities are ca. 50 km from each other. Crosses were made in the spring of 1985 and 1986 by transferring pollen from

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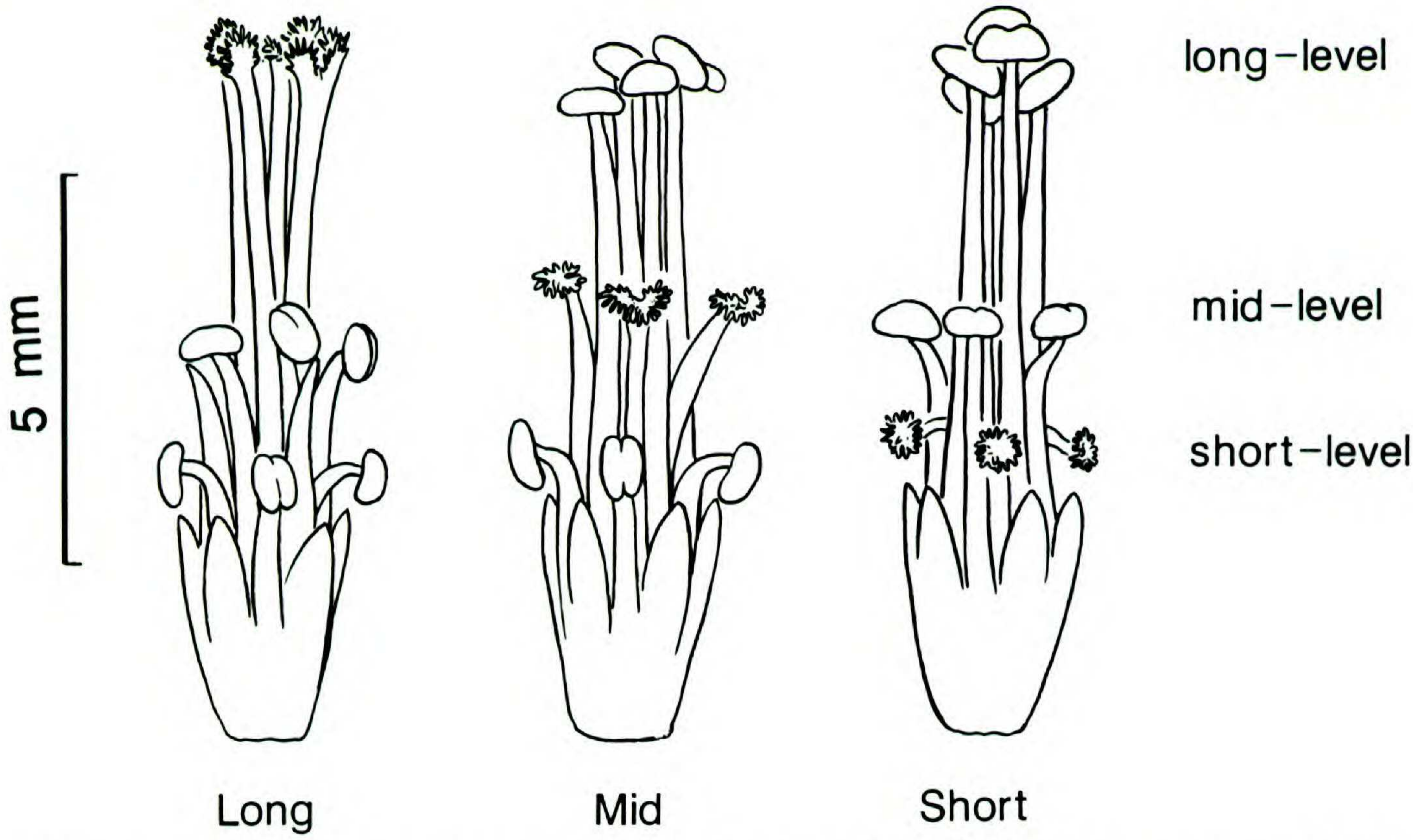

FIGURE 1. Illustrations of gynoecia and androecia of the three floral forms of *Oxalis pes-caprae*. The designation of each floral form is given below each illustration. The stigma and anther level designations used in the text are given to the right of the figure.

dehisced anthers to stigmas under insect-free conditions. Seed set was determined by collecting nearly mature capsules and counting the seeds released by them in seed packets. Pollen size for the three morphs was determined by mounting fresh grains in aniline blue-lactophenol and measuring them with an ocular micrometer; pollen stainability was determined by counting the number of stained grains in a sample of 100 mounted in this medium.

RESULTS

Chromosome numbers. Seven of the nine native populations examined were tetraploid ($n = 14$); two were diploid ($n = 7$; Table 1). Previously published reports, summarized in Table 2, indicate only tetraploidy and pentaploidy for the species.

Weediness. The two diploid populations and four of the tetraploid populations were growing under disturbed conditions such as roadsides, vineyards, or grainfields and were considered weedy; the others occurred in undisturbed conditions and were considered non-weedy (field data are lacking for one tetraploid population; Table 1).

Morph ratios. Morph ratios have been reported earlier (Ornduff, 1974). In two populations, morph ratios of Longs : Mids : Shorts were 1 : 1 : 1, both populations were considered, with some doubt, to be non-weedy. In four populations, morph ratios deviated from equality. In one of these, Longs were deficient; in another, Mids; in third, Shorts; and in the fourth, Longs and Mids were greatly outnumbered by Shorts. Three of these four populations were characterized as weedy (two for which chromosome counts are available were tetraploid).

Pollen size and stainability. Pollen size of diploids and tetraploids is trimorphic (Table 1). Pollen from the long-level anthers is largest, that from mid-level anthers is smaller, and that from short-level anthers is smallest. Pollen from anthers at equivalent levels in different morphs was generally of the same size within a population, but there were interpopulation differences in pollen size from equivalent anther sets. There was no correlation between differences in pollen size and in chromosome number of the diploids and tetraploids. Pollen from presumed pentaploids collected in California was extremely variable in size, even from an individual anther. Pollen stainability of diploids and tetraploids was variable, but was mostly over 60%. One Short tetraploid (*7038*, Table 1) had pollen with 12% and 25% stainability from its two sets of anthers. One

TABLE 1. Chromosome numbers, pollen size (μm), and pollen stainability in *Oxalis pes-caprae*.

Collection (author's or collector's field number)	Chromosome Number	Floral Morph	Origin of Pollen (size; standard deviation; stainability in percent) Long-level Anthers	Mid-level Anthers	Short-level Anthers
Cape Province, South Africa:					
7035: Worcester, riverbank, non-weedy	$n = 14$	Long		41.7; 1.83 (—)	35.8; 2.03 (—)
		Mid	45.7; 2.65 (—)		33.0; 2.05 (—)
7038: Klapmuts, roadside, weedy	$n = 14$	Short	54.1; 3.94 (12%)	41.2; 3.10 (25%)	
7041: Stellenbosch, vineyard, weedy	—	Short	49.5; 3.20 (66%)	41.8; 1.83 (69%)	
7096: Mamre Road Station, roadside, weedy	$n = 7$	Mid	46.4; 1.95 (99%)		33.7; 1.91 (98%)
7248: Gouda/Hermon, roadside, weedy	$n = 7$	Mid	42.7; 3.94 (—)		31.1; 2.20 (—)
7292: Hopefield/Malmesbury, field, non-weedy	—	Long		41.8; 1.83 (89%)	42.7; 4.84 (88%)
		Mid	50.1; 2.38 (86%)		38.9; 1.83 (78%)
		Short	50.4; 4.15 (93%)	41.5; 2.93 (98%)	
7301: Hopefield/Langebaan, field, non-weedy	$n = 14$	Long		39.0; 1.83 (78%)	30.3; 3.92 (79%)
		Mid	46.2; 1.68 (83%)		31.2; 1.54 (81%)
		Short	48.3; 5.73 (81%)	34.4; 2.03 (77%)	
8053: Calvinia, grain field, weedy	$n = 14$	Long		42.6; 2.42 (65%)	36.2; 2.52 (60%)
		Mid	52.2; 2.67 (64%)		35.0; 1.81 (40%)
		Short	47.2; 2.75 (74%)	38.0; 2.55 (73%)	
8055: Clanwilliam, veld, non-weedy	—	Long		38.2; 2.49 (75%)	32.6; 2.07 (68%)
		Mid	45.1; 2.89 (94%)		32.6; 2.30 (86%)
		Short	48.3; 2.87 (88%)	38.8; 2.83 (82%)	
Goldblatt *s.n.*: Rustenberg, near Stellenbosch, abandoned farmland	$n = 14$	Long		—; — (90%)	—; — (84%)
		Mid	—; — (66%)		—; — (62%)
		Short	—; — (70%)	—; — (81%)	
Goldblatt *s.n.*: Noordhoek, Cape Peninsula	$n = 14$	Long		—; — (93%)	—; — (88%)
		Mid	—; — (81%)		—; — (79%)
		Short	—; — (88%)	—; — (78%)	
Carlquist *s.n.*: Southwestern Cape Province	$n = 14$	Long		40.2; 2.46 (80%)	32.0; 1.93 (46%)
		Mid	50.8; 4.84 (92%)		34.5; 2.01 (96%)
		Short	49.7; 4.37 (67%)	41.8; 3.71 (63%)	
California, U.S.A.:					
Ornduff *s.n.*: Univ. California campus, Berkeley	—	Short	—; — (62%)	—; — (87%)	
	—	Short	—; — (58%)	—; — (51%)	
R. Dulberger *s.n.*: as above	—	Mid	—; — (49%)		—; — (58%)
R. Dulberger *s.n.*: College Ave., Berkeley	—	Mid	—; — (32%)		—; — (29%)

set of anthers of a few other collections (e.g., *8053* Mid, Carlquist *s.n.* Long) had pollen with low stainability. The two collections used in the crossing program had pollen stainabilities exceeding 62%. Presumed Short pentaploids collected in California had pollen stainabilities ranging from 29% to 87%. However, these pollen grains were variable in size, and the total number per anther was reduced compared with pollen production of diploids and tetraploids as estimated from the density of the pollen grains on the prepared slides.

TABLE 2. Published chromosome counts of *Oxalis pes-caprae*.

Chromosome Number	Locality of Population Examined	Reference
$2n = 28$	Cape Town, South Africa	Marks, 1956
	India (garden plants)	Mathew, 1958
	Madeira	Borgen, 1974
	South Australia	Oram in Symon, 1961
	Western Australia	Michael, 1964
$2n = 34$	India (Punjab)	Bir and Sidhu, 1978, 1979, 1980
		Sidhu, 1979
$2n =$ ca. 35	Italy	Vignoli, 1935, 1937
$2n = 35$	Unknown	Yamashita, 1935
	South Australia	Oram in Symon, 1961
	Western Australia	Franklin in Michael, 1964
	Cape Town, South Africa	Franklin in Michael, 1964

Compatibility relationships. Table 3 presents summarized results for various crosses using parent plants from Rustenberg and Noordhoek. Legitimate pollinations are those between anthers and stigmas at equivalent levels; all other pollinations are termed illegitimate (Darwin, 1877). Only 4% of the illegitimately pollinated flowers produced capsules compared with 87% of the legitimately pollinated flowers. The average number of seeds per pollination obtained from legitimate crosses was 15.9; from illegitimate ones it was 0.2. Self-pollinations produced 0–0.2 seeds per pollination. Intermorph, illegitimate pollinations produced 0–0.5 seeds per pollination. Intermorph, legitimate pollinations produced 13.0–20.9 seeds per pollination. The two populations used in the crossing program produced similar results for all classes of crosses. Seed production of Shorts following legitimate pollinations was slightly greater than that of the other two morphs. Both populations are tetraploid (Table 1).

DISCUSSION

In its native range in South Africa, *Oxalis pes-caprae* is represented by indigenous weedy and non-weedy races with a conventional tristylous floral morphology, including pollen-size trimorphism, and a fully developed trimorphic incompatibility system. Individuals are strongly self-incompatible and illegitimate cross-pollinations produce little or no seed. Legitimate pollinations produce considerable quantities of seed. The pentaploid form has been reported from that country as well (Michael, 1964), but it is unclear how common this race is there.

In South Africa, tetraploids are apparently more common than diploids, but weedy populations of both are known. Unequal morph ratios may be more common in weedy South African races than in non-weedy ones and are likely a consequence of vegetative propagation that is enhanced by physical disturbances of the habitat during agricultural or road-building activities. Morph ratios were found in which Longs, Mids, and Shorts, respectively, were deficient in numbers. Each unequal morph ratio differed from the others, with no morph(s) predominating overall. Thus there is no basis from observation of native races that accounts for the fact that the aggressive morph outside South Africa is short-styled.

Over most of its exotic range of distribution, *Oxalis pes-caprae* appears to be represented by a fairly sterile, pentaploid short-styled morph. As early as 1887, Hildebrand noted the prevalence of this short-styled form and its lack of seed set. Henslow (1891, 1910) also noted these features and described in some detail the means of vegetative reproduction later amplified by Galil (1968). Where introduced, the species is distributed by human agents and, in places, by other animals such as the mole-rat (Galil, 1967) or by birds (Young, 1958).

Despite the high level of pollen sterility of the short-styled pentaploid, it apparently reproduces occasionally by seed. This may result from self- or geitonogamous pollinations in populations where Shorts alone are represented. Illegitimate pollinations of sexual Shorts carried out in the present study produced small amounts of seed, which offers support for this suggestion. Vignoli (1937), in an embryological study of the species, noted rare sexual reproduction in the pentaploid but concluded also that apogamy may rarely occur in this race. Another line of evidence for

TABLE 3. Results of legitimate and illegitimate pollinations in two populations of *Oxalis pes-caprae*. The first figures summarize results for the Rustenberg population; the second figures summarize results for the Noordhoek population.

Style Length (♀ parent) × Anther Level/Style Length (♂ parent)[1]	Number of Flowers Pollinated	Number of Flowers Producing Capsules	Number of Seeds Obtained	Average Number of Seeds/ Pollination
Self-pollinations (all illegitimate):				
L × m/L selfed	18; 55	0; 6	0; 34	0; 0.6
L × s/L selfed	20; 12	0; 0	0; 0	0; 0
M × l/M selfed	38; 33	0; 1	0; 5	0; 0.2
M × s/M selfed	23; 35	0; 0	0; 0	0; 0
S × l/S selfed	18; 20	1; 0	1; 0	0.1; 0
S × m/S selfed	15; 12	0; 0	0; 0	0; 0
Intermorph, illegitimate pollinations:				
L × s/M	26; 20	0; 0	0; 0	0; 0
L × m/S	23; 13	1; 2	10; 7	0.4; 0.5
M × s/L	25; 29	0; 2	0; 8	0; 0.3
M × l/S	15; 26	0; 1	0; 1	0; 0
S × l/M	15; 21	0; 3	0; 9	0; 0.4
S × m/L	25; 30	1; 4	17; 6	0.7; 0.2
Intermorph, legitimate pollinations:				
L × l/M	18; 48	12; 41	279; 738	15.5; 15.4
L × l/S	27; 34	23; 30	430; 583	15.9; 17.2
M × m/L	33; 37	29; 38	413; 533	12.5; 14.4
M × m/S	54; 42	46; 34	797; 546	14.8; 13.0
S × s/L	13; 22	11; 20	222; 460	17.1; 20.9
S × s/M	11; 29	11; 25	255; 588	23.2; 20.1
Summary of all legitimate, illegitimate pollinations:				
Legitimate	156; 212	132; 188	2,396; 3,448	15.4; 16.3
Illegitimate	261; 306	3; 19	28; 70	0.1; 0.2

[1] Notation is as follows: "L × m/L selfed" means a long-styled flower (L) was self-pollinated with its own pollen from the mid-level (m) set of anthers. "L × s/M" means a long-styled flower was pollinated with pollen from the short-level stamens of a mid-styled flower. Figure 1 illustrates the three flower types.

occasional sexual reproduction of this form comes from the spontaneous appearance of Mids in otherwise short-styled populations as noted in southern Italy by Vignoli (1935) and in central California by Dulberger (pers. comm.). It also has been suggested that in Western Australia, where the pentaploid sometimes exists in mixed populations with tetraploids, hybridization between the two may occur (Michael, 1964). The variability of the species there would suggest frequent sexual reproduction (Peirce, 1973). Tetraploid populations with all three style lengths are known in South Australia (Symon, 1961) and Western Australia (Michael, 1964) and these presumably reproduce sexually as well as asexually. Tetraploids also have been reported from India and Madeira (Mathew, 1958; Borgen, 1974), but it is unknown whether these are composed of more than one morph.

Although the occurrence of tetraploids and pentaploids in Australia as a consequence of independent introductions is documented (Michael, 1964), it is possible to explain the origin of tetraploids within pentaploid populations by another mechanism. The pentaploids produce some viable pollen (Table 1; Vignoli, 1937; Bir & Sidhu, 1980; Michael, 1964). The illustrations and discussion of microsporogenesis in pentaploids by Vignoli (1935, 1937) indicate that pollen grains with $n = 14$ can be produced by such plants. If megasporogenesis also results in eggs with $n = 14$, fertilization of these eggs by diploid sperm would result in a tetraploid zygote.

The place and mode of origin of the weedy pentaploid race are uncertain. Lower (1963) suggested that it may have originated outside South Africa. A 5x chromosome count reported by Michael (1964) indicates that this race is present in South Africa, but it is apparently not common (although this remains to be documented). The origin of the weedy pentaploid race can only tentatively be reconstructed, but I believe that it likely occurred in South Africa. The simplest explanation for the origin of pentaploidy is that it resulted from the union of an unreduced gamete of a tetraploid plant with a haploid gamete from a diploid plant. Since diploids are unknown outside South Africa, this event must be assumed to have occurred in South Africa. Although diploids and tetraploids are not known to be sympatric in South Africa, they have been collected very near each other, so sympatry may occur. Weediness clearly preceded the occurrence of pentaploidy since sexual diploids and tetraploids are frequently aggressively weedy: they commonly occur in cultivated fields and along roadsides. Pentaploidy itself has not conferred weediness on the species. Likewise, prodigious means of vegetative reproduction occur in diploids and tetraploids with a fully developed sexual apparatus, so that the almost exclusively vegetative propagation characteristic of the pentaploid is a condition that likewise preceded the origin of pentaploidy.

Present evidence, although scanty, suggests that the largely sterile pentaploid race is less successful as a weed in South Africa than are the sexual diploids and tetraploids. Outside South Africa, however, the pentaploid seems to prevail, possibly as a consequence of its greater competitive success over sexual races under exotic conditions (as, for example, seems to be the case in Australia) or as a consequence of a series of coincidences of introduction that led to this race being more abundant than its apparent diploid and tetraploid precursors. There is a possibility that there are competitive differences among the chromosomal races of *Oxalis pes-caprae*, or among its morphs (as suggested by Peirce, 1973). The reduced pollen stainability of some field collections suggests the possibility that, in these, the sexual apparatus may be impaired and that asexual mechanisms are more important in their reproductive mode. Both suggestions merit study. Nevertheless, the sequence of events leading to the origin of pentaploidy and the routes of human-aided migration and introduction of this species to other continents will probably never be fully reconstructed.

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